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USPT	endonuclease?	6478	<u>L5</u>
USPT	((((435/6)!.CCLS.))	7106	<u>L4</u>
USPT	((435/199)!.CCLS.)	308	<u>L3</u>
USPT	mammalian adj flap adj endonuclease	1	<u>L2</u>
USPT	flap adj endonuclease	8	<u>L1</u>

WEST**Generate Collection****Search Results - Record(s) 1 through 8 of 8 returned.**☐ **1. Document ID: US 6090543 A**

L1: Entry 1 of 8

File: USPT

Jul 18, 2000

US-PAT-NO: 6090543

DOCUMENT-IDENTIFIER: US 6090543 A

TITLE: Cleavage of nucleic acids

DATE-ISSUED: July 18, 2000

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Prudent; James R.	Madison	WI	N/A	N/A
Hall; Jeff G.	Madison	WI	N/A	N/A
Lyamichev; Victor I.	Madison	WI	N/A	N/A
Brow; Mary Ann D.	Madison	WI	N/A	N/A
Dahlberg; James E.	Madison	WI	N/A	N/A

US-CL-CURRENT: 435/6; 435/91.5, 435/91.53

Full	Title	Citation	Front	Review	Classification	Date	Reference	Claims	KWIC	Draw Desc	Image
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☐ **2. Document ID: US 6090606 A**

L1: Entry 2 of 8

File: USPT

Jul 18, 2000

US-PAT-NO: 6090606

DOCUMENT-IDENTIFIER: US 6090606 A

TITLE: Cleavage agents

DATE-ISSUED: July 18, 2000

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Kaiser; Michael W.	Madison	WI	N/A	N/A
Lyamichev; Victor I.	Madison	WI	N/A	N/A
Lyamicheva; Natasha	Madison	WI	N/A	N/A

US-CL-CURRENT: 435/199; 435/320.1, 435/325, 536/23.7, 536/24.3

Full	Title	Citation	Front	Review	Classification	Date	Reference	Claims	KWIC	Draw Desc	Image
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☐ **3. Document ID: US 5994069 A**

L1: Entry 3 of 8

File: USPT

Nov 30, 1999

US-PAT-NO: 5994069

DOCUMENT-IDENTIFIER: US 5994069 A

TITLE: Detection of nucleic acids by multiple sequential invasive cleavages

DATE-ISSUED: November 30, 1999

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Hall; Jeff G.	Madison	WI	N/A	N/A
Lyamichev; Victor I.	Madison	WI	N/A	N/A
Mast; Andrea L.	Madison	WI	N/A	N/A
Brow; Mary Ann D.	Madison	WI	N/A	N/A

US-CL-CURRENT: 435/6; 435/91.5, 435/91.53

Full	Title	Citation	Front	Review	Classification	Date	Reference	Claims	KWIC	Draw Desc	Image
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☐ 4. Document ID: US 5985557 A

L1: Entry 4 of 8

File: USPT

Nov 16, 1999

US-PAT-NO: 5985557

DOCUMENT-IDENTIFIER: US 5985557 A

TITLE: Invasive cleavage of nucleic acids

DATE-ISSUED: November 16, 1999

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Prudent; James R.	Madison	WI	N/A	N/A
Hall; Jeff G.	Madison	WI	N/A	N/A
Lyamichev; Victor I.	Madison	WI	N/A	N/A
Brow; Mary Ann D.	Madison	WI	N/A	N/A
Dahlberg; James E.	Madison	WI	N/A	N/A

US-CL-CURRENT: 435/6; 536/23.1

Full	Title	Citation	Front	Review	Classification	Date	Reference	Claims	KWIC	Draw Desc	Image
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☐ 5. Document ID: US 5874283 A

L1: Entry 5 of 8

File: USPT

Feb 23, 1999

US-PAT-NO: 5874283
DOCUMENT-IDENTIFIER: US 5874283 A

TITLE: Mammalian flap-specific endonuclease

DATE-ISSUED: February 23, 1999

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Harrington; John Joseph	Shaker Heights	OH	44120	N/A
Hsieh; Chih-Lin	St. Louis	MO	63131	N/A
Lieber; Michael R.	St. Louis	MO	63131	N/A

US-CL-CURRENT: 435/252.3; 435/199, 435/252.33, 435/320.1, 435/69.1, 530/350,
536/23.2, 536/23.5

Full	Title	Citation	Front	Review	Classification	Date	Reference	Claims	KMOC	Draw Desc	Image
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☐ 6. Document ID: US 5871992 A

L1: Entry 6 of 8

File: USPT

Feb 16, 1999

US-PAT-NO: 5871992
DOCUMENT-IDENTIFIER: US 5871992 A

TITLE: Mammalian endonuclease III, and diagnostic and therapeutic uses thereof

DATE-ISSUED: February 16, 1999

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Teebor; George W.	New York	NY	N/A	N/A
Hilbert; Timothy P.	New York	NY	N/A	N/A

US-CL-CURRENT: 435/199

Full	Title	Citation	Front	Review	Classification	Date	Reference	Claims	KMOC	Draw Desc	Image
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☐ 7. Document ID: US 5843669 A

L1: Entry 7 of 8

File: USPT

Dec 1, 1998

US-PAT-NO: 5843669

DOCUMENT-IDENTIFIER: US 5843669 A

TITLE: Cleavage of nucleic acid acid using thermostable methoanococcus jannaschii
FEN-1 endonucleases

DATE-ISSUED: December 1, 1998

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Kaiser; Michael W.	Madison	WI	N/A	N/A
Lyamichev; Victor I.	Madison	WI	N/A	N/A
Lyamichev; Natasha	Madison	WI	N/A	N/A

US-CL-CURRENT: 435/6; 435/18, 435/183, 435/194, 435/195, 435/196, 435/4, 435/810,
435/822, 435/91.53, 436/94, 530/350

Full	Title	Citation	Front	Review	Classification	Date	Reference	Claims	KWIC	Draw Desc	Image
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☐ 8. Document ID: US 5719056 A

L1: Entry 8 of 8

File: USPT

Feb 17, 1998

US-PAT-NO: 5719056

DOCUMENT-IDENTIFIER: US 5719056 A

TITLE: Proteins from pyrococcus furiosus

DATE-ISSUED: February 17, 1998

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Brummet; Shauna R.	Wadsworth	OH	N/A	N/A
Robb; Frank T.	Silver Spring	MD	N/A	N/A
Borges; Kimberly M.	Coventry	CT	N/A	N/A
Hujer; Kristine M.	Cleveland	OH	N/A	N/A
Domke; Sally T.	Parma	OH	N/A	N/A

US-CL-CURRENT: 435/320.1; 435/191, 435/196, 435/212, 435/252.3, 435/69.1,
536/23.1, 536/23.2, 536/23.7

Full	Title	Citation	Front	Review	Classification	Date	Reference	Claims	KWIC	Draw Desc	Image
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Terms	Documents
flap adj endonuclease	8

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L1: Entry 1 of 8

File: USPT

Jul 18, 2000

US-PAT-NO: 6090543

DOCUMENT-IDENTIFIER: US 6090543 A

TITLE: Cleavage of nucleic acids

DATE-ISSUED: July 18, 2000

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Prudent; James R.	Madison	WI	N/A	N/A
Hall; Jeff G.	Madison	WI	N/A	N/A
Lyamichev; Victor I.	Madison	WI	N/A	N/A
Brow; Mary Ann D.	Madison	WI	N/A	N/A
Dahlberg; James E.	Madison	WI	N/A	N/A

ASSIGNEE-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY	TYPE CODE
Third Wave Technologies, Inc.	Madison	WI	N/A	N/A	02

APPL-NO: 8/ 759038

DATE FILED: December 2, 1996

PARENT-CASE:

This is a Continuation-In-Part of co-pending application Ser. No. 08/756,386, filed Nov. 26, 1996 which is a Continuation-In-Part of co-pending application Ser. No. 08/682,853, filed Jul. 12, 1996, which is a Continuation-In-Part of co-pending application Ser. No. 08/599,491, filed on Jan. 24, 1996 and a Divisional of co-pending application Ser. No. 08/758,314, filed Dec. 2, 1996.

INT-CL: [7] C12Q 1/68, C12P 19/34

US-CL-ISSUED: 435/6; 435/91.5, 435/91.53, 935/77, 935/78

US-CL-CURRENT: 435/6; 435/91.5, 435/91.53

FIELD-OF-SEARCH: 435/6, 435/91.2, 435/91.1, 435/91.5, 435/91.53, 536/24.3, 536/24.33, 935/6, 935/77, 935/78

REF-CITED:

U.S. PATENT DOCUMENTS

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<input type="checkbox"/>	<u>4511502</u>	April 1985	Builder et al.	260/112
<input type="checkbox"/>	<u>4511503</u>	April 1985	Olson et al.	260/112
<input type="checkbox"/>	<u>4512922</u>	April 1985	Jones et al.	260/112
<input type="checkbox"/>	<u>4518526</u>	May 1985	Olson	260/112
<input type="checkbox"/>	<u>4683195</u>	July 1987	Mullis et al.	435/6
<input type="checkbox"/>	<u>4683202</u>	July 1987	Mullis	435/91
<input type="checkbox"/>	<u>4775619</u>	October 1988	Urdea	435/6
<input type="checkbox"/>	<u>4876187</u>	October 1989	Duck et al.	435/6
<input type="checkbox"/>	<u>5011769</u>	April 1991	Duck et al.	435/6
<input type="checkbox"/>	<u>5108892</u>	April 1992	Burke et al.	435/6
<input type="checkbox"/>	<u>5118605</u>	June 1992	Urdea	435/6
<input type="checkbox"/>	<u>5210015</u>	May 1993	Gelfand et al.	435/6
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<input type="checkbox"/>	<u>5487972</u>	January 1996	Gelfand et al.	435/6
<input type="checkbox"/>	<u>5494810</u>	February 1996	Barany et al.	435/91.52
<input type="checkbox"/>	<u>5541311</u>	July 1996	Dahlberg et al.	536/23.7
<input type="checkbox"/>	<u>5545729</u>	August 1996	Goodchild et al.	536/24.5
<input type="checkbox"/>	<u>5614402</u>	March 1997	Dahlberg et al.	435/199

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FOREIGN-PAT-NO	PUBN-DATE	COUNTRY	US-CL
0 482 714 A1	October 1991	EPX	
89/09284	October 1989	WOX	
90/01069	February 1990	WOX	
91/09950	July 1991	WOX	
92/02638	February 1992	WOX	
92/06200	April 1992	WOX	
94/29482	December 1994	WOX	
95/14106	May 1995	WOX	
96/20287	July 1996	WOX	
96/40999	December 1996	WOX	

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ART-UNIT: 164

PRIMARY-EXAMINER: Jones; W. Gary

ASSISTANT-EXAMINER: Shoemaker; Debra

ATTY-AGENT-FIRM: Medlen & Carroll, LLP

ABSTRACT:

The present invention relates to means for the detection and characterization of nucleic acid sequences, as well as variations in nucleic acid sequences. The present invention also relates to methods for forming a nucleic acid cleavage structure on a target sequence and

cleaving the nucleic acid cleavage structure in a site-specific manner. The structure-specific nuclease activity of a variety of enzymes is used to cleave the target-dependent cleavage structure, thereby indicating the presence of specific nucleic acid sequences or specific variations thereof.

27 Claims, 102 Drawing figures

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L1: Entry 1 of 8

File: USPT

Jul 18, 2000

US-PAT-NO: 6090543

DOCUMENT-IDENTIFIER: US 6090543 A

TITLE: Cleavage of nucleic acids

DATE-ISSUED: July 18, 2000

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Prudent; James R.	Madison	WI	N/A	N/A
Hall; Jeff G.	Madison	WI	N/A	N/A
Lyamichev; Victor I.	Madison	WI	N/A	N/A
Brow; Mary Ann D.	Madison	WI	N/A	N/A
Dahlberg; James E.	Madison	WI	N/A	N/A

US-CL-CURRENT: 435/6; 435/91.5, 435/91.53

CLAIMS:

We claim:

1. A method of detecting the presence of a target nucleic acid molecule by detecting non-target cleavage products comprising:

a) providing:

i) a cleavage means,

ii) a source of target nucleic acid, said target nucleic acid having a first region, a second region, a third region and a fourth region, wherein said first region is located downstream from said second region, said second region is contiguous to and downstream from said third region and said third region is located adjacent to and downstream from said fourth region;

iii) a first oligonucleotide complementary to said fourth region of said target nucleic acid;

iv) second and third oligonucleotides having 3' and 5' portions, wherein said 3' portion of said second oligonucleotide contains a sequence complementary to said third region of said target nucleic acid and wherein said 5' portion of said second oligonucleotide and said 3' portion of said third oligonucleotide each contain sequence fully complementary to said second region of said target nucleic acid, and wherein said 5' portion of said third oligonucleotide contains sequence complementary to said first region of said target nucleic acid;

b) mixing said cleavage means, said target nucleic acid, said first oligonucleotide, said second oligonucleotide and said third oligonucleotide to create a reaction mixture under reaction conditions such that said first oligonucleotide is annealed to said fourth region of said target nucleic acid and wherein at least said 3' portion of said second oligonucleotide is annealed to said target nucleic acid and wherein at least said 5' portion of said third oligonucleotide is annealed to said target nucleic acid so as to create a cleavage structure and wherein cleavage of said cleavage structure occurs to generate non-target cleavage products, each non-target cleavage product having a 3'-hydroxyl group; and

c) detecting said non-target cleavage products.

2. The method of claim 1, wherein said target nucleic acid comprises single-stranded DNA.

3. The method of claim 1, wherein said target nucleic acid comprises double-stranded DNA and prior to step c), said reaction mixture is treated such that said double-stranded DNA is rendered substantially single-stranded.

4. The method of claim 1, wherein said target nucleic acid comprises RNA and wherein said first and second oligonucleotides comprise DNA.
5. The method of claim 1, wherein said cleavage means is a structure-specific nuclease.
6. The method of claim 5, wherein said structure-specific nuclease is a thermostable structure-specific endonuclease.
7. The method of claim 6, wherein said nuclease is encoded by a DNA sequence selected from the group consisting of SEQ ID NOS:1-3, 9, 10, 12, 21, 30, 31, 101, 106, 110, 114, 129, and 132.
8. The method of claim 1, wherein said detection of said non-target cleavage products comprises electrophoretic separation of the products of said reaction followed by visualization of said separated non-target cleavage products.
9. The method of claim 1, wherein one or more of said first, second, and said third oligonucleotides contain a dideoxynucleotide at the 3' terminus.
10. The method of claim 9, wherein said detecting said non-target cleavage products comprises:
 - a) incubating said non-target cleavage products with a template-independent polymerase and at least one labelled nucleoside triphosphate under conditions such that at least one labelled nucleotide is added to the 3'-hydroxyl group of said non-target cleavage products to generate labelled non-target cleavage products; and
 - b) detecting the presence of said labelled non-target cleavage products.
11. The method of claim 10, wherein said template-independent polymerase is selected from the group consisting of terminal deoxynucleotidyl transferase and poly A polymerase.
12. The method of claim 11, wherein said second oligonucleotide contains a 5' end label, said 5' end label being a different label than the label present upon said labelled nucleoside triphosphate.
13. The methods of claim 12, wherein said 5' end label is selected from the group consisting of biotin, fluorescein, tetrachlorofluorescein, hexachlorofluorescein, Cy3 amidite, Cy5 amidite and digoxigenin.
14. The method of claim 9, wherein following said detecting said non-target cleavage products comprises:
 - a) incubating said non-target cleavage products with a template-independent polymerase and at least one nucleoside triphosphate under conditions such that at least one nucleotide is added to the 3'-hydroxyl group of said non-target cleavage products to generate tailed non-target cleavage products; and
 - b) detecting the presence of said tailed non-target cleavage products.
15. The method of claim 14, wherein said second oligonucleotide contains a 5' end label.
16. A method of detecting the presence of a target nucleic acid molecule by detecting non-target cleavage products comprising:
 - a) providing:
 - i) a cleavage means,
 - ii) a source of target nucleic acid, said target nucleic acid having a first region, a second region and a third region, wherein said first region is downstream from said second region and wherein said second region is contiguous to and downstream from said third region;
 - iii) first and second oligonucleotides having 3' and 5' portions, wherein said 3' portion of said first oligonucleotide contains a sequence complementary to said third region of said target nucleic acid and wherein said 5' portion of said first oligonucleotide and said 3' portion of said second oligonucleotide each contain sequence fully complementary to said second region of said target nucleic acid, and wherein said 5' portion of said second oligonucleotide contains sequence complementary to said first region of said target nucleic acid;
 - b) mixing, in any order, said cleavage means, said target nucleic acid, said first oligonucleotide and said second oligonucleotide to create a reaction mixture under reaction conditions such that at least said 3' portion of said first oligonucleotide is annealed to said target nucleic acid and wherein at least said 5' portion of said second oligonucleotide is annealed to said target nucleic acid so as to create a cleavage structure and wherein cleavage of said cleavage structure occurs to generate non-target cleavage products, each non-target cleavage product having a 3'-hydroxyl group; and
 - c) detecting said non-target cleavage products.
17. The method of claim 16, wherein said cleavage means is a structure-specific nuclease.
18. The method of claim 16, wherein said second region of said target nucleic acid has a length between one to five nucleotides.
19. The method of claim 16, wherein at least said first oligonucleotide contains a

dideoxynucleotide at the 3' terminus.

20. The method of claim 16, wherein said detecting said non-target cleavage products comprises:

a) incubating said non-target cleavage products with a template-independent polymerase and at least one labelled nucleoside triphosphate under conditions such that at least one labelled nucleotide is added to the 3'-hydroxyl group of said non-target cleavage products to generate labelled non-target cleavage products; and

b) detecting the presence of said labelled non-target cleavage products.

21. The method of claim 16, wherein following said detecting said non-target cleavage products comprises:

a) incubating said non-target cleavage products with a template-independent polymerase and at least one nucleoside triphosphate under conditions such that at least one nucleotide is added to the 3'-hydroxyl group of said non-target cleavage products to generate tailed non-target cleavage products; and

b) detecting the presence of said tailed non-target cleavage products.

22. A method of detecting the presence of a target nucleic acid molecule by detecting non-target cleavage products comprising:

a) providing:

i) a cleavage means,

ii) a source of target nucleic acid, said target nucleic acid having a first region, a second region and a third region, wherein said first region is downstream from said second region and wherein said second region is contiguous to and downstream from said third region;

iii) a first and second oligonucleotide having 3' and 5' portion, wherein said 3' portion of said first oligonucleotide contains a sequence complementary to said third region of said target nucleic acid and wherein said 5' portion of said first oligonucleotide and said 3' portion of said second oligonucleotide each contain sequence fully complementary to said second region of said target nucleic acid, and wherein 5' portion of said second oligonucleotide contains sequence complementary to said first region of said target nucleic acid;

b) mixing, in any order, said cleavage means, said target nucleic acid, said first oligonucleotide and said second oligonucleotide to create a reaction mixture under reaction conditions such that at least said 3' portion of said first oligonucleotide is annealed to said target nucleic acid and wherein at least said 5' portion of said second oligonucleotide is annealed to said target nucleic acid so as to create a cleavage structure and wherein cleavage of said cleavage structure occurs to generate non-target cleavage products, each non-target cleavage product having a 3'-hydroxyl group; and

c) detecting said non-target cleavage products.

23. The method of claim 22, wherein said cleavage means is a structure-specific nuclease.

24. The method of claim 22, where said second region of said target nucleic acid has a length between one to five nucleotides.

25. The method of claim 22, wherein at least said first oligonucleotide contains a dideoxynucleotide at the 3' terminus.

26. The method of claim 22, wherein said detecting said non-target cleavage products comprises:

a) incubating said non-target cleavage products with a template-independent polymerase and at least one labelled nucleoside triphosphate under conditions such that at least one labelled nucleotide is added to the 3' -hydroxyl group of said non-target cleavage products to generate labelled non-target cleavage products; and

b) detecting the presence of said labelled non-target cleavage products.

27. The method of claim 22, wherein following said detecting said non-target cleavage products comprises:

a) incubating said non-target cleavage products with a template-independent polymerase and at least one nucleoside triphosphate under conditions such that at least one nucleotide is added to the 3'-hydroxyl group of said non-target cleavage products to generate tailed non-target cleavage products; and

b) detecting the presence of said tailed non-target cleavage products.

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L1: Entry 2 of 8

File: USPT

Jul 18, 2000

US-PAT-NO: 6090606

DOCUMENT-IDENTIFIER: US 6090606 A

TITLE: Cleavage agents

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INVENTOR-INFORMATION:

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US-CL-CURRENT: 435/199; 435/320.1, 435/325, 536/23.7, 536/24.3

CLAIMS:

We claim:

1. A thermostable structure-specific nuclease having an amino acid sequence selected from the group consisting of SEQ ID NOS:102, 107, 130 and 132.
2. The nuclease of claim 1, wherein said nuclease is encoded by a DNA sequence selected from the group consisting of SEQ ID NO:101, 106, 129 and 131.
3. A recombinant DNA vector comprising DNA having a nucleotide sequence encoding a structure-specific nuclease, said nucleotide sequence selected from the group consisting of SEQ ID NO:101, 106, 129 and 131.
4. A host cell transformed with the recombinant vector of claim 3.
5. The host cell of claim 4, wherein said host cell is an Escherichia coli cell.
6. A purified Pyrococcus woeisii FEN-1 endonuclease, wherein said Pyrococcus woeisii FEN-1 endonuclease is encoded by the plasmid pTrc99-PWFEN-1 (ACTT 207094).
7. The purified endonuclease of claim 6, wherein said endonuclease has a molecular weight of about 38.7 kilodaltons.
8. An isolated oligonucleotide comprising at least a portion of the plasmid pTrc99-PWFEN-1 (ACTT 207094), said portion encoding Pyrococcus woeisii FEN-1 endonuclease.
9. The isolated oligonucleotide of claim 8, wherein said oligonucleotide encoding said endonuclease is operably linked to a heterologous promoter.
10. The isolated oligonucleotide of claim 9, wherein said heterologous promoter is an inducible promoter.
11. The isolated oligonucleotide of claim 10, wherein said inducible promoter is selected from the group consisting of the .lambda.-P.sub.L promoter, the tac promoter, the trp promoter and the trc promoter.
12. A recombinant DNA vector comprising an isolated oligonucleotide, said oligonucleotide comprising at least a portion of the plasmid pTrc99-PWFEN-1 (ACTT 207094), said portion encoding Pyrococcus woeisii FEN-1 endonuclease.
13. A host cell transformed with the recombinant vector of claim 12.
14. The host cell of claim 13, wherein said host cell is an Escherichia coli cell.
15. An isolated oligonucleotide comprising a gene encoding a Pyrococcus woeisii FEN-1 endonuclease having a molecular weight of about 38.7 kilodaltons, wherein said Pyrococcus woeisii FEN-1 endonuclease comprises the plasmid pTrc-99FWFEN-1 (ACTT 207094).
16. The isolated oligonucleotide of claim 15, wherein said gene encoding a Pyrococcus woseii FEN-1 endonuclease is operably linked to a heterologous promoter.
17. The isolated oligonucleotide of claim 16, wherein said heterologous promoter

is an inducible promoter.

18. The isolated oligonucleotide of claim 17, wherein said inducible promoter is selected from the group consisting of the .lambda.-P.sub.L promoter, the tac promoter, the trp promoter and the trc promoter.

19. A recombinant DNA vector comprising DNA having a nucleotide sequence encoding a *Pyrococcus woesii* FEN-1 endonuclease having a molecular weight of about 38.7 kilodaltons, and wherein said *Pyrococcus woesii* FEN-1 endonuclease comprises the plasmid pTrc-99FWFEN-1 (ACTT 207094).

20. A host cell transformed with the recombinant vector of claim 19.

21. The host cell of claim 20, wherein said host cell is an *Escherichia coli* cell.

22. An isolated oligonucleotide encoding *Pyrococcus woesii* FEN-1 endonuclease, said oligonucleotide comprising a sequence fully complementary to at least 15 consecutive nucleotides of a sequence selected from the group consisting of SEQ ID NOS:116-119.

23. A recombinant DNA vector comprising the isolated oligonucleotide of claim 22.

24. A host cell transformed with the recombinant vector of claim 23.

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L1: Entry 6 of 8

File: USPT

Feb 16, 1999

US-PAT-NO: 5871992

DOCUMENT-IDENTIFIER: US 5871992 A

TITLE: Mammalian endonuclease III, and diagnostic and therapeutic uses thereof

DATE-ISSUED: February 16, 1999

INVENTOR-INFORMATION:

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US-CL-CURRENT: 435/199

CLAIMS:

What is claimed is:

1. A mammalian endonuclease III purified greater than about 500-fold, which endonuclease III demonstrates pyrimidine hydrate DNA-glycosylase activity, thymine glycol DNA-glycosylase activity, and AP lyase activity, and reductively cross links with a thymine glycol containing oligodeoxynucleotide.
2. The endonuclease III of claim 1, wherein the endonuclease is purified greater than about 5000-fold.
3. The endonuclease III of claim 1, wherein the endonuclease in 100 mM NaCl elutes from a 1 ml single stranded-DNA-cellulose chromatography column eluted with a 12.5 ml gradient of 100 to 600 mM NaCl at 0.2 ml/min in about fractions 12-18.
4. The endonuclease III of claim 2 which elutes in about fractions 15-17.
5. The endonuclease III of claim 1 which has an apparent molecular weight of 29 kDa as determined by gel filtration.
6. The endonuclease III of claim 1 which has a predominant molecular weight of 31 kDa as determined by SDS-PAGE analysis.
7. The endonuclease III of claim 1 which has a partial amino acid sequence selected from the group consisting of SEQ ID NO:25, SEQ ID NO:26, SEQ ID NO:27, SEQ ID NO:28, SEQ ID NO:6, and SEQ ID NO:20.
8. The endonuclease III of claim 1 which has an amino acid sequence selected from the group consisting of bovine endonuclease III, human endonuclease III, and rat endonuclease III.
9. An endonuclease III having an amino acid sequence corresponding to SEQ ID NO:2.